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FROG'S BLADDER TO WATER

A DISSERTATION SUBMITTED TO THE  
GRADUATE FACULTY IN CANDIDACY FOR  
THE DEGREE OF DOCTOR OF PHILOSOPHY

DEPARTMENT OF ANATOMY

By  
WILLIAM BROOKS STEEN

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## ON THE PERMEABILITY OF THE FROG'S BLADDER TO WATER

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In several recent investigations on the secretion of urine in the frog the assumption is apparent that the urine contained in the frog's bladder represents in quantity and composition that actually delivered to the urogenital duct by the kidney tubules. This situation has given rise to certain discrepancies in the results of renal investigation which require explanation. The consistent results of a group of investigators, including Przylecki ('21),<sup>1</sup> Bainbridge, Menzies, and Collins ('14), Adolph ('27), Crane ('27), and others, indicate that the urine collected from the frog's bladder, though varying in concentration with different conditions of hydration of the animal, nevertheless is never hypertonic to the blood of the same animal, and is frequently hypotonic. Nussbaum ('78), on the other hand, reports a distinct increase of concentration in the tubules visible from the ventral surface of the pronephros of Triton, and this observation has recently been confirmed by Bieter and Hirschfelder ('24), Richards and Barnwell ('27), and by Bensley and Steen ('28). The latter report that in kidneys deprived of their glomerular circulation by ligation of the renal arteries the kidney continues to eliminate indigo-carmin and that this is concentrated in the distal convoluted tubule to such an extent that the dye fills, in crystalline form, the distended lumen of the tubule.

In the discussion of her results Crane attempts to explain the apparent incapacity of the frog's kidney to concentrate

<sup>1</sup> Work reported by J. K. Parnas ('21).

its product on the basis that the kidney of the frog, unlike that of the mammal, lacks the loop of Henle as a component part of the renal tubule, and presumes that this segment of the tubule is responsible for the phenomena of concentration. In none of these investigations is a suspicion apparent that the concentration of the urine might be modified in the urogenital duct or in the bladder itself.

In one of his earlier papers Nussbaum made the extraordinary observation that when frogs were removed from water to air they ceased to secrete urine, and this statement was recently confirmed by Adolph ('27) in the following words: "When frogs were removed from water and placed in a moist atmosphere they ceased altogether to form urine." Since Adolph was collecting urine from the bladder, this statement simply means that under these conditions the bladder did not contain an appreciable quantity of fluid.

The results of all of the recorded experiments as to volume of urine in the urinary bladder and as to its concentration in comparison with that of the blood would be equally well explained if it were shown that the bladder was freely permeable to water and that water was absorbed under conditions of dehydration until osmotic equilibrium with the blood was approached or attained.

If the bladder wall is thus freely permeable to water under normal conditions, it follows that the urinary bladder of these animals plays a much more important rôle than that of a mere temporary receptacle of urine, for since the urine of the frog as it reaches the bladder, in the normal animal immersed in water, is a highly dilute solution, the bladder may also serve as a source of water to tide over periods when the animals are not in a position to receive it by the usual route of absorption through the skin.

Accordingly, I have performed a number of experiments to ascertain whether under conditions of dehydration the water in the bladder could be resorbed into the blood.

It has been shown by Adolph ('27) that in frogs with the cloacal aperture closed by a ligature and immersed in water



the increase in weight from hour to hour approximates the weight of urine which accumulates in the bladder. A first series of experiments was conducted to test this conclusion.

A series of four frogs was prepared as follows: The bladders were first evacuated through the cloaca, and the animals were wiped dry with a cloth and weighed. Then a suture was applied around the cloacal opening and the animals were placed in water. At the end of the indicated lapse of time they were reweighed, and the bladder was then opened and the urine collected in a graduate and measured. The results of this experiment are indicated in table 1.

This series shows that in frogs whose cloacal opening has been closed by a ligature, living in water, the increase in body weight parallels closely the accumulation of water in the bladder.

TABLE 1

| <i>Frog<br/>no.</i> | <i>Weight<br/>Grams</i> | <i>Time in water<br/>Minutes</i> | <i>Final weight<br/>Grams</i> | <i>Gain<br/>Grams</i> | <i>Urine<br/>Cc.</i> |
|---------------------|-------------------------|----------------------------------|-------------------------------|-----------------------|----------------------|
| 1                   | 63.7                    | 655                              | 70.75                         | 7.05                  | 5.5                  |
| 2                   | 49.7                    | 660                              | 57.3                          | 7.6                   | 6.7                  |
| 3                   | 51.7                    | 677                              | 58.6                          | 6.9                   | 7.0                  |
| 4                   | 37.3                    | 611                              | 45.55                         | 8.25                  | 8.5                  |

Taking advantage of this fact, it was now possible to prepare a series of frogs in which the content of urine in the bladder was approximately known, and to submit them to conditions of dehydration. The frogs were prepared as before, weighed, placed in water at room temperature for a period of approximately six hours, weighed again to ascertain the amount of urine in the bladder, then placed for about seven hours in a 1 per cent solution of sodium chloride. At the end of the period they were weighed again to ascertain the loss in weight, and the bladder was opened and the urine measured. The results of this experiment are recorded in table 2.

Table 2 shows that there was an initial gain of weight of the animals in tap-water indicating an accumulation of urine in the bladder of from 4.2 to 6.5 cc. in the period elapsed. During the succeeding period in 1 per cent salt solution, urine

practically disappeared from the bladder, and the weight of the frog in some instances was further reduced by the loss of more water through the skin than was recovered from the bladder.

This experiment was repeated, with the exception that the dehydration was accomplished by placing the animals in a dry chamber instead of immersing them in 1 per cent salt solution. Examination of the results recorded in table 3 shows that in these cases also, the accumulated urine was practically completely resorbed from the bladder during the period of dehydration.

TABLE 2

| <i>Frog<br/>no.</i> | <i>Weight</i> | <i>Time in water</i> | <i>Weight</i> | <i>Gain weight</i> | <i>Time in 1%<br/>salt solution</i> | <i>Weight</i> | <i>Loss weight</i> | <i>Urine</i>              |
|---------------------|---------------|----------------------|---------------|--------------------|-------------------------------------|---------------|--------------------|---------------------------|
|                     | <i>Grams</i>  | <i>Minutes</i>       | <i>Grams</i>  | <i>Grams</i>       | <i>Minutes</i>                      | <i>Grams</i>  | <i>Grams</i>       | <i>Cc.</i>                |
| 1                   | 65            | 357                  | 69.7          | 4.7                | 431                                 | 61.5          | 8.2                | 1 drop                    |
| 2                   | 58.8          | 345                  | 64.5          | 5.7                | 434                                 | 61.           | 3.5                | .4, with<br>some<br>feces |
| 3                   | 58.5          | 336                  | 65.           | 6.5                | 438                                 | 59.7          | 5.3                | .25                       |
| 4                   | 55.8          | 319                  | 60.           | 4.2                | 441                                 | 56.           | 4.                 | .25                       |

TABLE 3

| Frog no. | Weight<br>Grams | Time in water<br>Minutes | Weight<br>Grams | Gain weight<br>Grams | Time in dry chamber<br>Minutes | Weight<br>Grams | Loss weight<br>Grams | Urine<br>Cc.            |
|----------|-----------------|--------------------------|-----------------|----------------------|--------------------------------|-----------------|----------------------|-------------------------|
| 1        | 34.8            | 474                      | 38.35           | 3.55                 | 1154                           | 31.9            | 7.45                 | Feces with possible .25 |
| 2        | 48.2            | 478                      | 51.6            | 3.4                  | 416                            | 50.39           | 1.21                 | Repeated below          |
|          | 50.39           | 737                      | 58.1            | 7.71                 | 1079                           | 41.39           | 16.71                | 0                       |

A fourth series of experiments was then run to determine whether the diminution of urine in the bladder during dehydration was really due to resorption from the bladder or to the escape of the bladder contents into the intestinal tract to be there resorbed. In this series the animals were first opened under urethane anaesthesia and a ligature applied around the intestine just in front of the place where the bladder and urogenital ducts enter the cloaca. The abdomen was then closed by sutures and the animals allowed to recover from the anaesthetic. Then they were wiped dry, weighed, and a ligature was applied around the cloacal orifice. They



were then placed in water for several hours to allow the accumulation of urine in the bladders. The weight was then recorded, and the frogs were transferred to a dry chamber for dehydration, weighed again, and the bladder examined for contents. Frogs 2, 3, and 4 of this series were opened and the bladder found to contain urine after the sojourn in tap-water, then sewed up and transferred to the dry chamber. The abdomen of frog 1 was not reopened, but the frog was treated as in the previous experiment. The results are recorded in table 4.

TABLE 4

| <i>Frog no.</i> | <i>Weight Grams</i> | <i>Time in water Minutes</i> | <i>Weight Grams</i> | <i>Gain weight Grams</i> | <i>Time in dry chamber Minutes</i> | <i>Weight Grams</i> | <i>Loss weight Grams</i> | <i>Urine Cc.</i> |
|-----------------|---------------------|------------------------------|---------------------|--------------------------|------------------------------------|---------------------|--------------------------|------------------|
| 1               | 41                  | 572                          | 47.2                | 6.2                      | 1053                               | 41                  | 6.2                      | 0                |
| 2               | 51.8                | 620                          | 62                  | 10.2                     | 887                                | 56.1                | 5.9                      | 0                |
| 3               | 48                  | 597                          | 55                  | 7                        | 838                                | 52.3                | 2.7                      | 0                |
| 4               | 45.7                | 585                          | 53.6                | 7.9                      | 707                                | 50.7                | 2.9                      | 0                |

In all four of these animals the urine disappeared from the bladder during the period of dehydration. In frog 1 the loss of weight during dehydration was exactly equal to the increase of weight during the preliminary period in tap-water. In frogs 2, 3, and 4, on the contrary, the loss in weight in the dehydration period was less than the gain in weight during the hydration period—a fact which is probably to be explained by a change in the hydration capacity of the frog's tissues, due to the additional manipulation.

I wish to express my deep gratitude to Dr. R. R. Bensley for his help in this work.

## CONCLUSIONS

These experiments show that the increase of weight of frogs whose cloacal aperture is tied, preventing the escape of urine from the bladder, is mainly due to the accumulation of urine in the bladder.

They show, also, that under conditions of dehydration the contents of the bladder may be almost completely resorbed through the bladder wall.

It follows from these experiments and the observations of other investigators that the product of the kidneys as delivered to the bladder is modified by many factors:

1. The kidney product may be dilute or concentrated, varying with different conditions of hydration of the animal.
2. The urine in the bladder may be further concentrated by the reabsorption of water through the bladder wall.
3. The reabsorption occurs under conditions in which the change in the degree of concentration of the bladder urine makes it approach osmotic equilibrium with the blood.

## LITERATURE CITED

- ADOLPH, E. F. 1927 The excretion of water by the kidneys of frogs. *Am. Jour. Physiol.*, vol. 81, pp. 315-324.
- BAINBRIDGE, F. A., MENZIES, J. A., AND COLLINS, S. H. 1914 The formation of urine in the frog. *Jour. Physiol.*, vol. 48, pp. 233-243.
- BENSLEY, R. R., AND STEEN, W. B. 1928 The function of the differentiated segments of the uriniferous tubule. *Am. Jour. Anat.*, vol. 41, pp. 75-96.
- BIETER, R. N., AND HIRSCHFELDER, A. D. 1924 The excretion of dyes and other substances in the frog's kidney and its bearing upon the theories of renal secretion. *Am. Jour. Physiol.*, vol. 68, pp. 326-337.
- CRANE, M. M. 1927 Observations on the function of the frog's kidney. *Am. Jour. Physiol.*, vol. 81, pp. 232-243.
- NUSSEBAUM, M. 1878 Fortgesetzte Untersuchungen über die Sekretion der Niere. *Pflüger's Arch. f. d. ges. Physiol.*, Bd. 17, S. 580-594.
- PARNAS, J. K. 1921 Neue Untersuchungen über den Wasserhaushalt der Frosche. *Biochemische Zeitschrift*, Bd. 114, S. 1-11.
- RICHARDS, A. N., AND BARNWELL, J. B. 1927 Experiments concerning the question of secretion of phenolsulphonephthalein of renal tubules. *Proc. Roy. Soc., B*, vol. 102, pp. 72-90.







